PERSPECTIVE ARTICLE

Nanobiomaterial Applications in Orthopedics

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ABSTRACT: Advancements in nanobiotechnology are revolutionizing our capability to understand biological intricacies and resolve biological and medical problems by developing subtle biomimetic techniques. Nanocomposites and nanostructured materials are believed to play a pivotal role in orthopedic research since bone itself is a typical example of a nanocomposite. This article reviews current strategies using nanobiomaterials to improve current orthopedic materials and examines their applications in bone tissue engineering. Preliminary investigations support the potential of nanobiomaterials in orthopedic applications; however, significant advancements are necessary to achieve clinical use. Overall, current trends in nanobiotechnology foreshadow a bright future through the use of nanobiomaterials in the orthopedic domain. © 2006 Orthopaedic Research Society. Published by Wiley Periodicals, Inc. J Orthop Res 25:11–22, 2007

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INTRODUCTION

Due to the aging of our population, the market for orthopedic implants is growing at a rapid rate. Each year, more than 600,000 joint replacements are performed in the USA alone with an estimated worldwide cost in excess of 3 billion dollars. Metals are the most common choice for total bone replacement or implant fixations. The excellent mechanical properties of metals meet the necessary requirements for load-bearing bone applications. However, both metal and polymeric implants may fail due to stress-shielding, joint loosening due to wear, and limited compatibility with bone tissue. Failed implants require several challenging revision surgeries that drastically increase cost and recovery time.

Tissue engineering emerged as a promising alternative for the reconstitution of lost or damaged organs and tissues, circumventing the complications associated with traditional transplants.[1] Tissue engineers attempt to repair or regenerate damaged tissue by using engineered tissue substitutes that can sustain functionality during regeneration and eventually integrate with the host tissue. Initially, many synthetic structures were designed to impart bulk properties to the construct, such as adequate mechanical strength and sufficient transport properties for cell infiltration and tissue organization. Although many of these structures bore close resemblances to the

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macroscopic properties of native tissue, the constructs failed prior to full healing.[2,3]

The success of both the orthopedic implant and the tissue engineered construct is highly dependent on the selected biomaterial. One of the key factors identified in the failure of both types of implants was insufficient tissue regeneration around the biomaterial immediately after implantation. This has been attributed to poor surface interaction of biomaterials with the host tissue. It is known that the introduction of an implant into a living organism causes specific reactions in the biological environment. The biomolecules and cells together with the intrinsic properties of the chosen biomaterials determine the biocompatibility and longevity of the implants. Since the interaction of those biomolecules and cells with the biomaterial surface is a vital element in the evaluation of the biomaterial, biomaterial scientists have reexamined the pertinent host–cell interactions in order to design materials that facilitate favorable interactions and enhance tissue regeneration. Ultimately, improved symbiosis should result in an accelerated healing time, an increase in implant longevity, and a reduction in the necessity for revision surgery.

Research has shown that all living systems are governed by molecular behavior at nanometer scales. The molecular building blocks of life—proteins, nucleic acids, lipids, and carbohydrates—are examples of materials that possess unique properties determined by the size, folding, and patterns at the nanoscale. Specifically, the organization of cells and the corresponding tissue properties are found to be highly dependent on the structure of the extracellular matrix (ECM). The ECM has a complex hierarchical structure with spatial and temporal levels of organization that span several orders of magnitude (nm to cm scale). For these reasons, cells in our body are predisposed to interact with nanostructured surfaces.[4] Yet, most of the current macro- or microfabrication techniques are unable to recreate sophisticated structures that could mimic the subtleties of the ECM. Recent paradigm shifts from these fabrication techniques to nanoscience-enabled techniques have significantly enhanced our ability to design and develop better tissue substitutes. The unique feature of this nanotechnological approach is that it enables the consideration of spatial and temporal levels of material organization in order to develop appropriate hierarchical structures. Indeed, there is research evidence that a biomaterial substrate composed of nanometer-scale components is biologically preferred.[5,6] Nanometer structural components are thus being considered as promising biomaterials.

Although various definitions are attached to the word "nanomaterial" by different experts, the commonly accepted concept refers to materials with a nano-sized topography or composed of nano-sized building components. Examples include materials with a basic structural unit in the range 1–100 nm (nanostructured), crystalline solids with grain sizes 1–100 nm (nanocrystals), individual layers or multilayer surface coatings in the range 1–100 nm (nanocoatings), extremely fine powders with an average particle size in the range 1–100 nm (nanoparticles), and fibers with a diameter in the range 1–100 nm (nanofibers). This perspective article seeks to demonstrate the potential of nanobiomaterials to improve biological applications pertinent to orthopedics. Although nanobiomaterials have extensive applications in all areas of orthopedic research, we focused our perspective on current strategies using nanobiomaterials in bone research.

CELLULAR RECOGNITION OF NANOSCALE STRUCTURE

A common objective in orthopedic research is the design of biomaterials that support cell and tissue growth. An emerging area of research has combined traditional design with active modulation of cellular activities. In native tissues, nanoscale protein interactions are crucial to controlling cell functions such as proliferation, migration, and ECM production.[7] Protein adsorption characteristics are in turn dependent on the surface features of the implanted biomaterials (roughness, charge, chemistry, wettability).[8] The particulate or fiber size of the biomaterial influences these surface properties and the corresponding protein interactions. Recent reports have demonstrated that the unique properties of nanobiomaterials provide advantageous interactions with the proteins that control cellular function.[9,10]

Nanobiomaterials have an increased number of atoms and crystal grains at their surfaces and possess a higher surface area to volume ratio than conventional microscale biomaterials. These differences in surface topography alter the corresponding surface energy for protein adsorption. Literature reports indicate that cumulative adsorption of proteins from bodily fluids is significantly higher on smaller, nanometer, grain size materials.[9,10] In particular, the interaction of four proteins known to enhance osteoblast functions—fibronectin, vitronectin, laminin, and collagen—was shown to increase greatly on nanobiomater-

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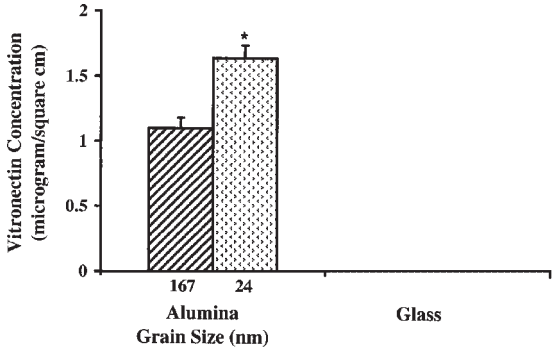

Figure 1. Enhanced vitronectin adsorption on nanophase alumina. Calcium-mediated vitronectin adsorption was investigated on borosilicate glass (reference material), conventional alumina (167-nm grain size), and nanophase alumina (24-nm grain size) alumina substrates. Compared to conventional alumina and borosilicate glass, calcium-mediated vitronectin adsorption was significantly ($p = 0.01$) greater on nanophase alumina. Calcium-mediated vitronectin adsorption was not detected on borosilicate glass (reference material) in the present study. Values are means ± SEM; $n = 3$; *$p < 0.01$ (compared to conventional grain size alumina). Reprinted with permission from Webster et al., Mechanisms of enhanced osteoblast adhesion on nanophase alumina involve vitronectin, Tissue Engineering 7:291–301. Copyright 2001 Mary Ann Liebert, Inc.

ials as compared to conventional (micron-sized grains) materials. Webster et al. correlated enhanced vitronectin adsorption, comformation, and bioactivity to the increased osteoblast adhesion on nanophase alumina (see Fig. 1).[9]

In addition to modulating protein adsorption, strategies to control the density, clustering, and orientation of cell signaling epitopes of adsorbed proteins are being explored.[7] A novel adaptation of the standard surface-enhanced Raman Scattering technique provided evidence of increased unfolding of proteins adsorbed on nanophase versus conventional ceramics.[9] These conformational changes promote availability of specific cell-adhesive epitopes that increase osteoblast adhesion and function. For example, osteoblasts have been shown to adhere to select amino acid sequences (such as Arginine-Glycine-Aspartic Acid or RGD) in proteins adsorbed onto biomaterial surfaces. Since osteoblast adhesion on a newly implanted orthopedic surface is imperative for those cells to synthesize bone, optimizing the initial protein adsorption events is integral to implant success. ECM production and mineralization are also extremely important when designing orthopedic implants. Thus, surfaces that promote cell adhesion should also be optimized for ECM production and subsequent mineralization. It is becoming increasingly evident that the key to modulating these critical protein interactions, and subsequent cellular behavior and tissue regeneration, lies in utilizing nanobiomaterials.

NANOBIOMATERIALS AS BONE IMPLANTS

The potential for modulating cellular behavior with nanobiomaterials has generated a landslide of research in the orthopedic domain. An exhaustive review of all the applications within orthopedics is beyond the scope of a single article. We focused our perspective on nanobiomaterials in bone research as an example of the current design strategies.

Biomimetic Nanocomposites

The use of nanotechnology to tailor orthopedic implant surfaces arises from the nanoscale structure of the ECM. As a three-dimensional architecture, the organic and inorganic components of bone ECM form an environment replete with informational cues for the cell types in bone tissue. The inorganic component of bone ECM is comprised of nanoscale calcium phosphate (CaP) crystallites similar to hydroxyapatite. In view of the native components of bone, CaP materials are logical choices as biomaterials. Indeed, CaP ceramics show good biological properties as they have the capacity to form a chemically bonded interface with bone.[11] However, the mechanical properties of bulk synthetic CaP materials are insufficient for their use at load-bearing, orthopedic sites. Consequently, CaPs are mostly used as coatings on metallic (mostly titanium and its alloys) bulk materials. In a recent study, tantalum porous scaffolds were coated with nanometer and conventional hydroxyapatite particles.[12] Compared to scaffolds coated with conventional grain size hydroxyapatite, in vivo osteointegration of porous tantalum coated with nanophase hydroxyapatite (see Fig. 2) was observed after 6 weeks of implantation into rat calvaria.

Several techniques are available for the deposition of CaP-coatings on metals, including plasma spraying, biomimetic deposition, laser deposition, ion beam deposition, radiofrequent (RF) magnetron sputter deposition, and Electrostatic Spray Deposition (ESD). Although plasma spraying is most frequently used for the deposition of CaP-coatings on orthopedic and dental implants, limitations of this technique include the lack of controlling coating structure, the relatively low cohesion within the thick coatings (~50 μm), and the limited bond strength with the metallic implant substrate.[13] Biomimetic deposition of CaP-coatings

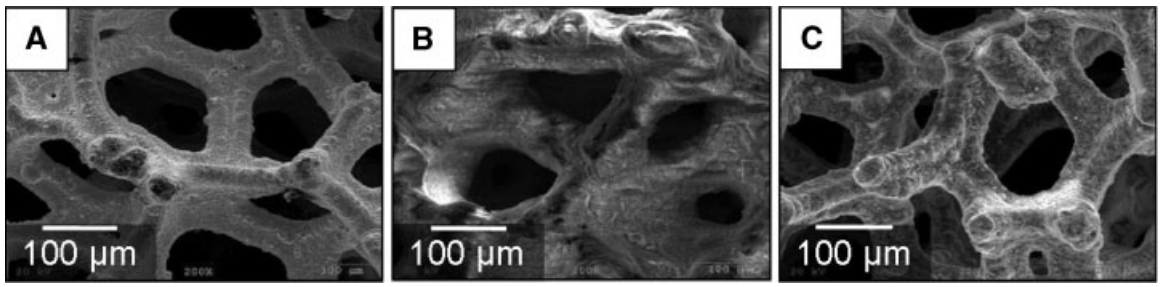

Figure 2. Scanning electron micrographs of (A) uncoated tantalum and (B) tantalum coatings of either micron grain size hydroxyapatite or (C) nanophase grain size hydroxyapatite. Energy-dispersive spectroscopy confirmed the presence of hydroxyapatite chemistries on the tantalum coatings. Bar = 100 μm. Reprinted with permission from Sato et al., Increased osseointegration for tantalum scaffolds coated with nanophase compared to conventional hydroxyapatite, International Journal of Nanomedicine, in press. Copyright 2006 Dove Medical Press.

results in the deposition of CaP crystals with nanoscale dimensions.[14,15] An interesting aspect of the biomimetic process of CaP deposition is that it can be combined with the deposition of biologically active compounds.[16,17] Through this co-precipitation process, the resulting CaP coatings do not only introduce bioactivity but can also actively influence cellular processes and reduce bacterial infections.

The last decade of research has focused on the deposition of nanoscale CaP-coatings utilizing RF magnetron sputter deposition, ESD and Electrostatic Self-Assembly (ESA). RF magnetron sputtering has successfully deposited CaP-coatings with nanometer thickness on metallic[18] as well as polymeric materials.[19,20] Even more interesting from a nanotechnological perspective are the CaP-coatings derived using ESD. This technique has the advantage that the compositional and morphological properties can be tailored by choosing appropriate combinations of deposition parameters.[21] Consequently, ESD allows the fabrication of porous CaP-coatings, which have a larger surface area available for interactions with adsorbed proteins. Interactions of these surface-bound proteins with cell receptors can then modulate cellular behavior. Additionally, this increased surface area enlarges the potential of porous CaP-coatings for drug delivery. Current experiments are focused on the effects of these porous CaP-coatings in vitro and in vivo, and on their drug delivery capacity. Another application of the ESD-technique is the spraying of suspensions containing nano-sized CaP particles.[22,23] ESA is another powerful technique to modify biomaterial surfaces. This technique employs oppositely-charged polyelectrolytes to form a multilayered structure.[24,25] Due to the variety of polyelectrolytes available (natural as well as synthetic), desirable surface properties can be tailored to specific applications.

Nanostructured Biomaterials

Decreased ceramic grain sizes have been correlated to increased bone cell function in the literature. Specifically, compared to conventional (micron grain size) ceramic formulations, ceramics made separately from spherical nanometer particles of alumina, titania, and hydroxyapatite enhanced in vitro adhesion of osteoblasts. Increased osteoblast functions were also observed at ceramic spherical grain sizes (or consequently, surface spherical bumps) below 60 nm.[26] Thus, evidence was provided that the ability of nanophase ceramics to promote bone cell function was indeed limited to below 100 nm. Studies further reported enhanced in vitro calcium deposition by osteoblasts as well as increased functions of osteoclasts (bone-resorbing cells) on nanophase ceramics.[5,27] Specifically, deposition of calcium by osteoblasts on nanophase alumina and titania was greater than on respective conventional ceramic formulations (see Fig. 3). Osteoclast synthesis of tartrate-resistant acid phosphatase and subsequent formation of resorption pits was also increased on nanophase as compared to conventional ceramics. Another design parameter to consider for orthopedic nanomaterials is particle aspect ratio. Consolidated substrates formulated from nanofibrous alumina (diameter: 2 nm, length $>$50 nm) demonstrated significantly increased in vitro osteoblast functions in comparison with similar alumina substrates formulated from nanospherical particles.[28] This result suggests that not only is the grain size of bone important to mimic in

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nanophase ceramics, but its fibrous aspect ratio may also be important to emulate in synthetic materials.

In addition to ceramics, nanophase metals (such as titanium, Ti6Al4V, and CoCr alloys) and polymers have also demonstrated the novel properties which promoted bone cell functions on ceramics. Specifically, in vitro bone cell adhesion was markedly greater on nanophase compared to conventional materials. In a disease-specific study, researchers have reported increased in vitro osteoblast functions on nanophase compared to conventional selenium (a metalloid).[29] Since selenium has been reported to have certain "anti-cancer" properties, such results highlight the potential use of nanophase selenium in implants for those with bone cancer. Although the metal nanotopographies in this study were created by consolidating nanoparticles, there are other ways to create nanometer roughness on metal surfaces. Compared to unanodized titanium, greater in vitro osteoblast adhesion and mineral deposition was observed on titanium anodized to possess nanometer tubes.[10] The same trends have also been observed for anodized aluminum.[30] Similarly, increased in vitro osteoblast functions were measured on poly(lactic-co-glycolic acid) (PLGA) cast from nanophase compared to conventional titania.[31] These results suggest that the proactive surface roughness of nanophase materials may be transferable to polymers to promote orthopedic implant efficacy.

NANOBIOMATERIALS AS BONE TISSUE ENGINEERING SCAFFOLDS

Tissue engineering is emerging as a potential alternative to current therapies to repair bone defects. Bone tissue engineering using nanobiomaterials is at the infant stage and growing at an exponential rate. Recent developments in modifying existing conventional materials to possess nanoscale features and increase new bone synthesis offer exciting opportunities in bone tissue engineering. In addition to increasing bioactivity and tissue integration, nanophase materials can also be used to improve the mechanical properties of scaffolds to match that of the native tissue.

Nanocomposite Constructs

Nanocomposite bone grafts made of hydroxyapatite-collagen exhibit some features of natural bone in composition and structure. As a scaffold material, hydroxyapatite (HA) facilitates greater osteoconduction and related functions than conventional materials.[32] However, it is not osteoinductive and its biodegradability is relatively slow. To circumvent these drawbacks, biodegradable polymers (i.e., collagen) can be employed to make a composite in conjunction with osteogenic potential cells and osteoinductive growth factors. Based on the experimental results, this tissue-engineered HA-collagen nanocomposite system seems to be very promising in engineering bone tissues. In subsequent studies, the nanoHA/collagen/osteoblast system was developed in conjunction with poly(lactic acid).[33] The construct was observed to support cellular adhesion, proliferation, and migration. In vivo efficacy was evaluated in a rabbit model in a subsequent study. The results showed integration of the segmental defect and evidence of new bone tissue formation. The system has great potential for the clinical repair of large bony defects according to the principles of bone tissue engineering. Although in vitro and in vivo evidence strongly supports the use of nanobiomaterials as a new kind of bone graft, further preclinical studies are required to realize their full potential in bone and other orthopedic applications.

Studies in the literature have shown significant increases in bone cell functions when nanophase (compared to conventional) ceramic particles were incorporated into polymer composites.[34] Specifically, up to three times more osteoblasts adhered to PLGA when it contained nanophase compared to conventional titania at the same weight ratio and porosity.[35] Moreover, significantly greater in vitro osteoblast functions leading to mineral deposition was observed on carbon fibers with nanometer compared to conventional dimensions.[36,37] Such novel cytocompatibility properties of carbon nanofibers/nanotubes have been translated to polymer composites; specifically, increased in vitro osteoblast adhesion was observed in polyurethane composites with greater weight percentages of nanometer compared to conventional carbon fibers (5%–75% weight percent).[38] In fact, there was a preferred alignment of the osteoblasts and subsequent deposition of calcium containing mineral along the carbon nanofibers present on the surfaces of the polymer.[39]

In addition to modulating cellular function to promote tissue regeneration, researchers have also focused on the challenge of designing bone tissue engineering scaffolds that mimic the unique mechanical properties of bone. Nanophase reinforcement has the potential of improving current materials to achieve mechanical strength comparable to the native tissue. Recently, carbon

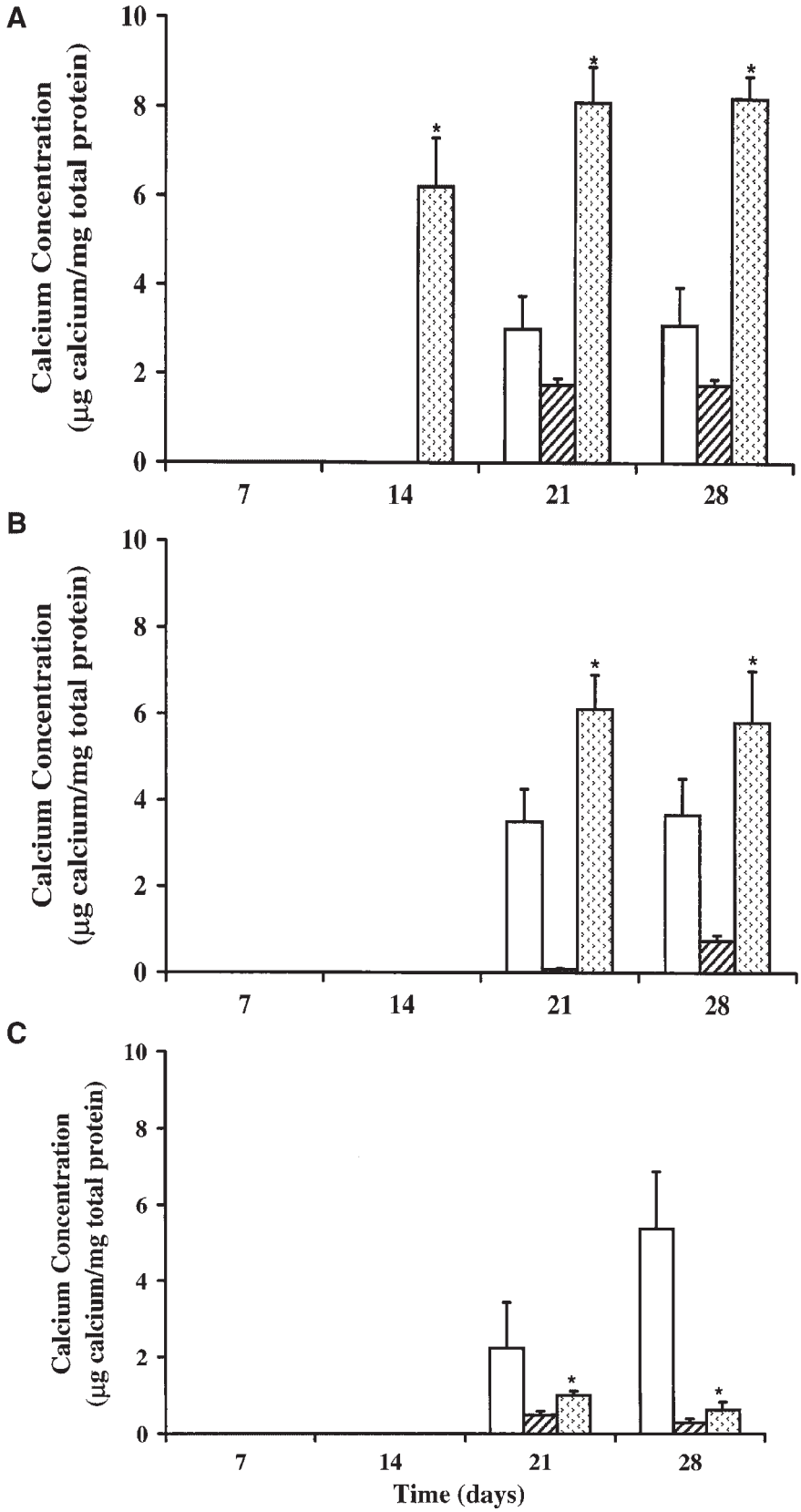

Figure 3.

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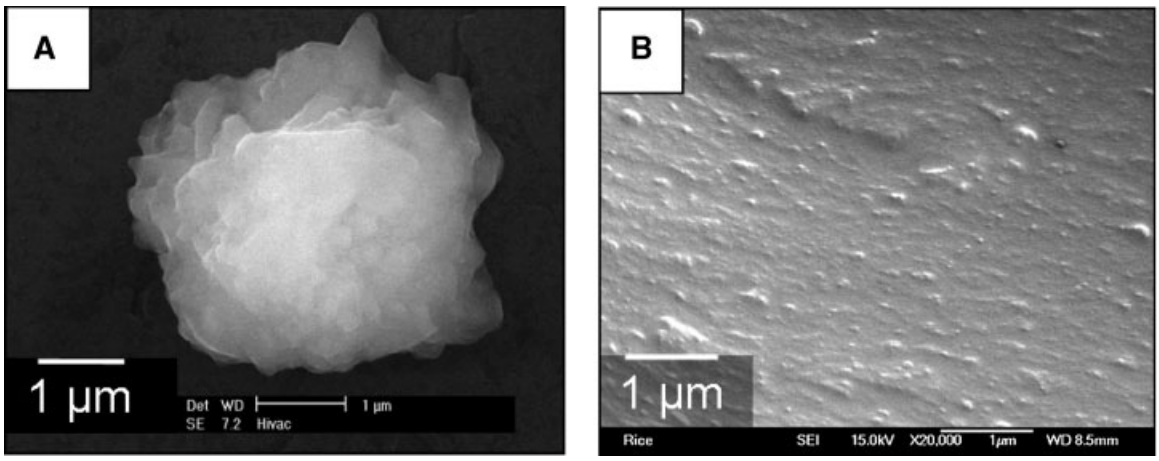

Figure 4. Scanning electron micrographs of fracture planes of nanocomposite samples after flexural testing (1 wt % loading): (A) unmodified boehmite crystals in polymer; (B) acryloyl undecanoic amino acid-alumoxane nanocomposite (hybrid). Bar = 1 μm. Reprinted with permission from Horch et al., Nanoreinforcement of poly(propylene fumarate)-based networks with surface modified alumoxane nanoparticles for bone tissue engineering, Biomacromolecules 5:1990–1998. Copyright 2004 American Chemical Society.

nanotubes and alumoxane nanoparticles have been examined as reinforcing fillers for biodegradable polymers.[40–42] The effect of a filler on mechanical properties is dependent on the size, shape, and dispersion of the filler. In addition, the interaction between the filler and the organic matrix can also impact the level of reinforcement. Therefore, optimal performance is achieved when the small particles are uniformly dispersed throughout the polymer and interact strongly with the organic matrix.[41] Both nanophase additives have a tendency to aggregate, losing their nanoscale size and corresponding properties.[41,42] Therefore, surface modification is necessary to improve miscibility such that a uniform dispersion may be achieved.

In a recent study, surface-modification of carboxylate alumoxane nanoparticles was used to improve optimal dispersion in the biodegradable polymer poly(propylene fumarate)/poly(propylene fumarate)-diacrylate (see Fig. 4).[41] The fine dispersion of nanoparticles and increased interaction between polymer chains and nanoparticles resulted in a threefold increase in flexural modulus with no significant loss of compressive or flexural strength.[41] Modification of single-walled carbon nanotubes (SWNTs) was also examined to improve their dispersion in poly(propylene fumarate). Although improvements in compressive and flexural mechanical properties were observed at low concentrations of SWNT, higher concentrations resulted in significant SWNT aggregation regardless of surfactant or functionalization.[42] These results indicate that SWNTs can be used to improve the mechanical properties of a biodegradable polymer; however, improved dispersion of individual SWNTs at higher additive concentrations is

Figure 3. Extracellular calcium deposited by osteoblasts cultured on the following substrates: (A) (open bars) borosilicate glass (reference material), (hatched bars) conventional alumina (39-nm grain size), and (stippled bars) nanophase alumina (24-nm grain size); (B) (open bars) borosilicate glass, (hatched bars) conventional titania (4,520-nm grain size), and (stippled bars) □ nanophase titania (39-nm grain size); and (C) (open bars) borosilicate glass, (hatched bars) conventional hydroxyapatite (179-nm grain size), and (stippled bars) nanophase hydroxyapatite (67-nm grain size). Extracellular calcium concentration (μg calcium/mg protein) was determined after 7, 14, 21, and 28 days. Values are means ± SEM; $n = 3$; *$p < 0.01$ (compared to respective conventional grain size ceramic). Reprinted with permission from Webster et al., Enhanced functions of osteoblasts on nanophase ceramics, Biomaterials 21:1803–1810. Copyright 2000 Elsevier.

necessary to fully recognize the potential of these reinforcing nanofillers in bone tissue engineering scaffolds.

Nanofiber Scaffolds

Nanofiber matrices have shown tremendous promise as tissue engineering scaffolds for bone regeneration. Nanofibers are particularly suitable for use as scaffolding components compared to nanoparticles, due to their continuous structure. The biomimetic environment of the nanofiber matrix affects cell–cell and cell–matrix interactions for favorable cell behavior. The advantages of a scaffold composed of ultra-fine, continuous fibers are high porosity, variable pore-size distribution, high surface-to-volume ratio, and most importantly, morphological similarity to natural ECM.[43] In addition, both in vitro[44] and in vivo[45] results have shown that mesenchymal stem cells undergo osteogenic differentiation with the support of nanofibrous scaffolds. Human bone marrow stromal cells were found to adhere and proliferate well on a polymeric nanofiber scaffold. In fact, the cells were found to crosslink the nanofibers in the matrix and integrate with the surrounding fibers to form a three-dimensional cellular network. Even though the pore diameter of a nanofiber matrix is small, it has been found to present a dynamic architecture to cells in culture. Cells migrate through the matrix by optimizing the pore size via pushing the surrounding fibers aside, as nanoscale fibers offer very little resistance to amoeboid movement of the cell.

A number of techniques, such as phase separation,[46] self-assembly,[47] and electrospinning,[43] have been developed based on different physical principles to fabricate nanofibrous scaffolds with unique properties. Among these techniques, electrospinning technology has become popular for the fabrication of tissue engineering scaffolds in recent years, because of the growing interest in nanotechnology and the unique properties and relative ease of fabricating scaffolds using this process. The versatility of the electrospinning process to develop scaffolds for tissue engineering is exceptional. Typically, an electrospun nanofiber matrix shows a porosity of more than 90% and pore diameter range up to 100 μm. Another unique feature of the electrospinning process is the feasibility to develop nanofiber scaffolds having varying sizes and shapes. Scanning electron micrographs of electrospun chitosan-based nanofibers with adherent osteoblast-like cells are shown in Figure 5.[48]

A large number of polymeric biomaterials have been electrospun into nanofibrous scaffolds, including nonbiodegradable and biodegradable polymers, with the latter consisting of both natural and synthetic polymers. Nonbiodegradable polymers, such as polyurethane[49] and polyesterurethane,[50] can be utilized to engineer tissues requiring substantial mechanical stability, such as ligament or muscle, but their long-lasting nature is likely to interfere with tissue turnover and remodeling. Therefore, more attention has been devoted to biodegradable polymers in tissue engineering. Polymer biodegradation, by the combined effect of enzymatic and hydrolytic activities, generates

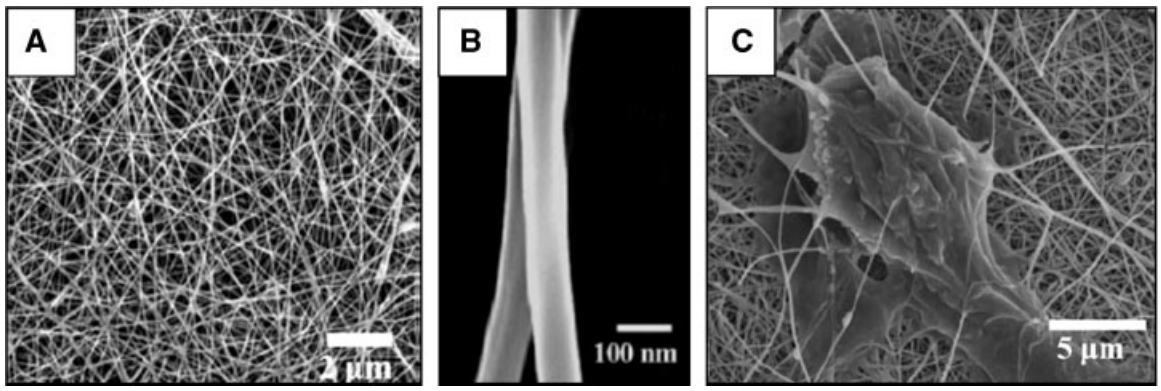

Figure 5. Scanning electron micrographs of (A) electrospun nanofibrous mesh made of chitosan/poly(ethylene oxide) with weight ratio of 90/10 (bar = 2 μm); (B) high-magnification of the nanofibers shown in (A) (bar = 100 nm); and (C) osteoblast-like cells (MG-63) seeded on the nanofibrous mesh after 5 days culture (bar = 5 μm). Reprinted with permission from Bhattarai et al., Electrospun chitosan-based nanofibers and their cellular compatibility, Biomaterials 26:6176–6184. Copyright 2005 Elsevier.

space within the scaffold that facilitates cellular processes, such as proliferation and the deposition of newly synthesized ECM. To date, more than 100 different biodegradable polymers have been successfully electrospun, and over 30 of them have been used for a variety of tissue-engineered applications.[51] Among the natural polymers, collagen, gelatin, elastin, silk fibroin, fibrinogen, hyaluronan, and chitosan have recently been fabricated into three-dimensional, nanofibrous scaffolds for orthopedic applications.

The poly(α-hydroxy ester) polymer family is the most commonly used synthetic biodegradable polymers for nanofiber production and have shown promise for orthopedic applications.[52] For example, poly(ϵ-caprolactone) and poly(L-lactic acid) based nanofibrous scaffolds have been successfully used for cell-based engineering of cartilage and bone tissues in vitro. In addition to poly(α-hydroxy esters), bioresorbable polyphosphazenes have also been examined as candidates for nanofiber materials.[53] Bioresorbable polyphosphazenes form a unique class of polymer for biomedical applications due to excellent biocompatibility, near neutral degradation products and synthetic flexibility, which allows for the development of polymers having unique chemical, physical, and biological properties.[53,54] A recent study demonstrated the ability to develop nanofibers from polyphosphazenes having appropriate side groups to nucleate and deposit hydroxyapatite.[55] In addition, literature reports indicate the feasibility of developing composite nanofibers by encapsulating nanohydroxyapatite particles within polyphosphazene nanofibers or electrospraying nanohydroxyapatite on electrospun nanofibers to develop scaffolds having better osteoconductivity and osteointegration.[56]

In summary, electrospinning has developed into one of the most elegant techniques to develop nanostructured scaffolds that could closely mimic the dimension of collagen fibrils in the ECM. Studies so far have demonstrated the versatility of the process to control the structure and morphology of the fiber matrices and their favorable interactions with cells for tissue organization. However, the biological processes that govern cell–cell and cell–matrix interactions controlled by various biochemical cues present in the natural ECM are just as important as these structural features. Current studies focused on developing hierarchical structures with spatially presented biological cues from bioresorbable electrospun nanofibers could lead to the development of ideal scaffolds for tissue engineering applications.

NANOBIOMATERIALS IN OTHER ORTHOPEDIC APPLICATIONS

Many of the same nanobiomaterials design strategies used in bone research have also been applied to other musculoskeletal tissues. For example, nanofiber scaffolds have been used in tissue engineering constructs of cartilage, tendon, and ligament. Recent findings indicate that the biological activities of chondrocytes[57] and mesenchymal stem cells[58] are crucially dependent on the dimensionality of the extracellular scaffolds. Due to the morphologic similarity to the collagen fibers in the extracellular matrix of natural tissue, nanofiber scaffolds may prove to be a biologically preferred scaffold/substrate for proliferation and phenotype maintenance of chondrocytes and chondrogenic differentiation of mesenchymal stem cells. Indeed, electrospun nanofiber scaffolds were shown to support chondrocytic phenotype of fetal bovine chondrocytes and chondrogenic induction and maintenance of TGF-β1 treated MSCs.[40,59,60] Nanofiber scaffolds have also been implemented in ligament and tendon reconstruction research.[61]

Similarly, nanostructured biomaterials have shown great promise as scaffold materials for cartilage tissue engineering. Enhanced chondrocyte adhesion was observed on biomaterials with nanoscale topography. There are several strategies being utilized to generate nanostructured biomaterials for cartilage research. For example, nanoadditives such as nanophase titania have been used in biomaterial composites.[62] Manipulation of the surface roughness by chemical degradation has also been used to generate nanoscale features.[63] These studies lay the foundation for increased research utilizing nanostructured biomaterials at the bone/cartilage interface.

KEY CHALLENGES AND CRITICAL ISSUES

Although preliminary investigations seem to support the impact of nanobiomaterials in orthopedic research, significant advancements are necessary to realize their full potential in clinical use. To move to the next developmental phase of nanobiomaterial science, it is critical to understand the cellular and molecular basis governing the interaction between nanostructure and cells. Substantial research efforts are required to address the following key challenges and critical issues:

- Consistency of processing technologies of nanobiomaterials

- Optimization of structure and properties mimicking natural bone
- Matching the strength of nanobiomaterials-based constructs with those of the natural bone in order to provide a uniform distribution of stresses (load sharing)
- Optimizing bioresorption of nanobiomaterials without comprising mechanical properties
- Identifying cell-specific nanobiomaterials
- Understanding molecular mechanisms of cell–nanobiomaterial interactions
- Improving angiogenesis within the nanobiomaterials system
- Assessing the inflammatory response to nanobiomaterials to validate their biosafety.

In particular, the risks to human health and environment must not be overlooked. Many issues relating to safe fabrication of nanobiomaterials still need to be addressed. For example, small nanoparticles may enter the human body through pores and may accumulate in the cells of the respiratory or other organ systems (when becoming dislodged through wear debris), and the health effects are yet to be largely known. This could happen during commercial-scale processing of the nanoparticles as well as through the use of these materials as implants. Continuous monitoring is necessary to assess the potential effects of newly designed and fabricated nanomaterials.

CONCLUDING REMARKS

The scientific developments reported above do not exhaust the current global beehive of research efforts on the biological potentials of nanobiomaterials as implants. The application of nanotechnology to biomaterial science is a new frontier in orthopedic research. Nanotechnology enables the development of new systems that mimic the complex, hierarchical structure of the native tissue to great effect. The preliminary investigations indicate that these strategies have great potential to improve current orthopedic biomaterials and in the development of new tissue engineering scaffolds. However, significant advancements are necessary to realize the full potential of nanobiomaterials in clinical use. Overall, current trends in nanotechnology foreshadow a bright future through the use of nanobiomaterials in the orthopedic domain.

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